

An Efficient Synthesis of *trans*-*N*-{2-[4-(2-Methoxyphenyl)piperazinyl]ethyl}-*N*-(2-pyridyl)-*N*-(4-tosyloxymethylcyclohexane)carboxamide, A Precursor of [^{18}F]Mefway for Imaging 5-HT_{1A} Receptor

Jae Yong Choi,^{†,‡} Chul Hoon Kim,[‡] Jung Young Kim,^{§,#} Hyun-Joon Ha,[#] and Young Hoon Ryu^{†,*}

[†]Department of Nuclear Medicine, Yonsei University College of Medicine, Gangnam Severance Hospital, 612 Eonjuro, kangnam-gu, Seoul 135-720, Korea. *E-mail: ryuyh@yuhs.ac

[‡]Department of Pharmacology, Brain Research Institute, Brain Korea 21 Project for Medical Science, Yonsei University College of Medicine, Seoul 120-752, Korea

[§]Molecular Imaging Research Center, Korea Institute of Radiological and Medical Sciences, 215-3, Gongneung-dong, Nowon-gu, Seoul 139-706, Korea

[#]Department of Chemistry and Protein Research Center for Bio-Industry, Hankuk University of Foreign Studies, Yongin, Kyunggi-Do 449-791, Korea

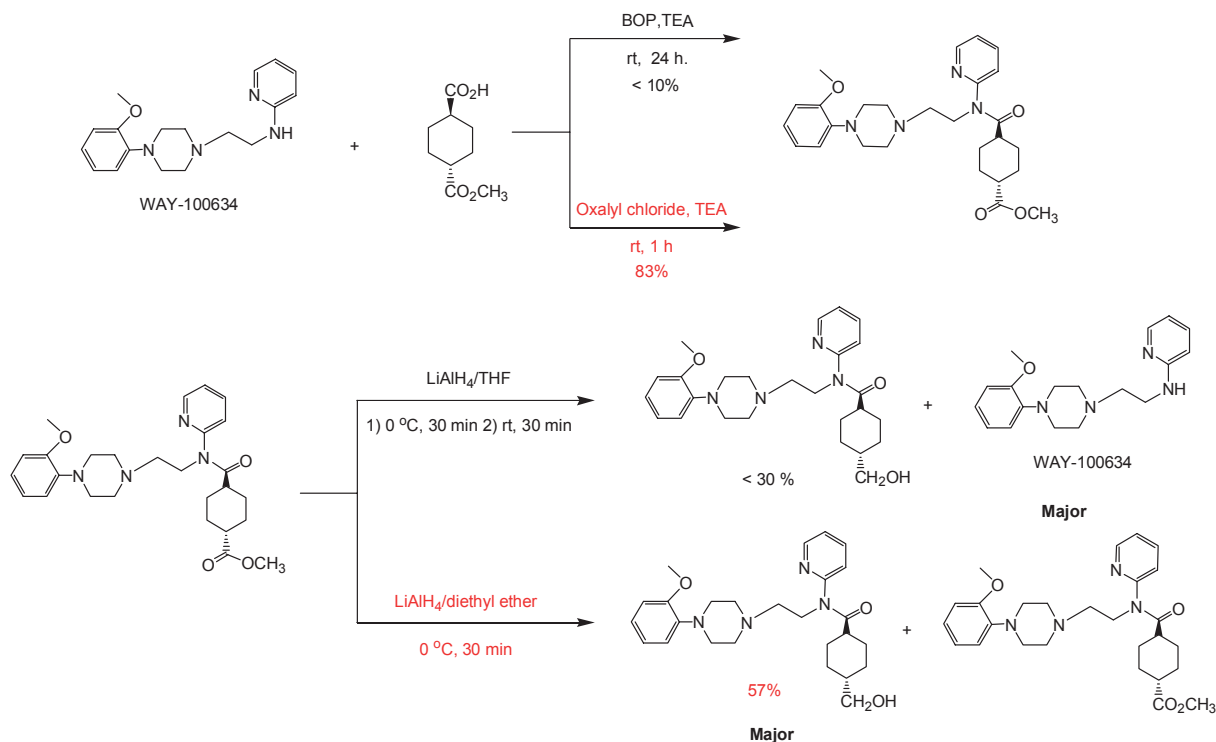
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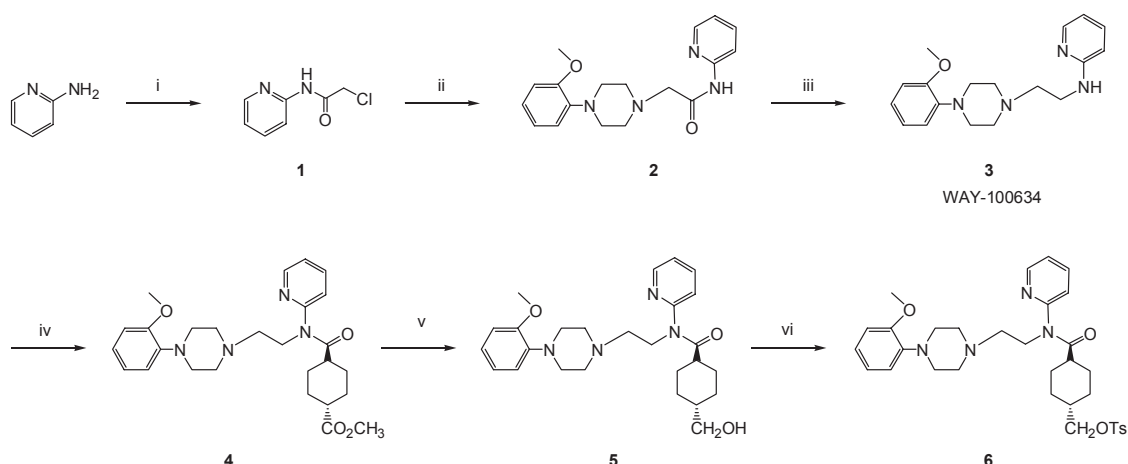
The serotonin (5-HT) system, one of the most important neurotransmitter systems, has been classified into seven subtypes (5-HT₁₋₇). Among these subfamilies, 5-HT_{1A} receptors in the central nervous system are strongly implicated in psychiatric disorders such as depression, anxiety and schizophrenia.^{1,2} Thus molecular imaging agents for the 5-HT_{1A} have been intensively studied since the past decade.^{3,4} Positron emission tomography (PET) as a non-invasive imaging technique with a high-sensitivity (10^{-9} – 10^{-12} M) and quantitative property, provides the means to visualize receptor densities in living system.^{5,6} PET

can play an important role both in assessing the neuropsychiatric disorders and in therapies with already developed pharmaceuticals.^{7,8,9}

It is known that only high affinity agonists bind to receptors while antagonists' binding is relatively insensitive to their affinity.^{10,11} Thereby, a number of antagonist tracers have been developed as PET imaging agents on the basis of the WAY-100635 which is a selective antagonist for 5-HT_{1A} receptors at both somatodendritic and postsynaptic receptor sites.¹²⁻¹⁶ Structural analogues were designed to have stability to metabolism



Scheme 1. Key steps in synthesis of the precursor



Scheme 2. Synthesis of Mefway precursor. Reagents and conditions: i) chloroacetyl chloride, TEA, CH₂Cl₂, rt, 79%, ii) 1-(2-methoxyphenyl)piperazine, K₂CO₃, NaI, DMF, 80 °C, 84%, iii) LiAlH₄, THF, rt, 76%, iv) oxalylchloride, *trans*-4-carbomethoxycyclohexane-1-carboxylic acid, CH₂Cl₂, rt, 83%, v) LiAlH₄, Et₂O, 0 °C, 57%, vi) Ts₂O, TEA, CH₂Cl₂, rt, 75%.

with high affinity, and selectivity for the 5-HT_{1A}. The most important drawback of these compounds is the significant defluorination that causes low-quality images due to contamination of ¹⁸F-fluoride ion in the skull. Recently, Neil Saigal *et al.* reported [¹⁸F]Mefway ((*N*-2-{2-[4-(2-methoxyphenyl)piperazinyl]ethyl}-*N*-(2-pyridyl)-*N*-(4-¹⁸F-fluoro-methylcyclohexane)carboxamide) as a potent PET agent for 5-HT_{1A} receptors. This compound had not only high target-to-non target ratios in receptor rich regions but also little defluorination.¹⁷

However, synthesis of its precursor, *trans*-*N*-2-[2-[4-(2-methoxyphenyl)piperazinyl]ethyl]-*N*-(2-pyridyl)-*N*-(4-tosyloxymethylcyclohexane)carboxamide, prior to the fluorination was quite inefficient with the low overall yield for the development of radiopharmaceutical due to the significant breakdown of amide bond, i.e., up to 70% of starting substrate, during the reduction of carbomethoxy group toward WAY-100635 derivative. Thereby, an efficient synthesis of *trans*-[¹⁸F]Mefway precursor is needed which we would like to describe with adaptation of acid chloride-assisted coupling reaction and an improved reduction method.

WAY-100634 (**3**) was prepared as previously described.⁸ Reaction of 2-aminopyridine with chloroacetyl chloride at room temperature provided the 2-(chloroacetyl)amidopyridine (**1**) as a intermediate. The treatment of the intermediate with 1-(2-methoxyphenyl)piperazines in DMF at 80 °C in the presence of K₂CO₃ and NaI gave the corresponding *N*-2-[2-{4-(2-methoxyphenyl)-1-piperazinyl}ethyl]amidopyridine (**2**), which is subsequently reduced to desired product **3**, using LiAlH₄ in THF at room temperature.

The WAY-100634 was coupled to *trans*-4-carbomethoxycyclohexanecarbonyl chloride in dichloromethane in the presence of triethylamine at room temperature, yielding *trans*-*N*-2-[2-[4-(2-methoxyphenyl)piperazinyl]ethyl]-*N*-(2-pyridyl)-*N*-(4-carboxymethylcyclohexane)carboxamide (**4**). In this reaction, we used oxalyl chloride as a coupling agent instead of benzotriazole-1-yloxytris-(dimethylamino)-phosphonium hexafluoro-phosphate (BOP) to enhance reactivity of carboxylic

acid. This method improved the reaction yield up to 83% higher than previously reported method using BOP.¹⁷ Specific reduction of carbomethoxy group in **4** was conducted in LiAlH₄ in Et₂O at 0 °C to provide *trans*-*N*-2-[2-[4-(2-methoxyphenyl)piperazinyl]ethyl]-*N*-(2-pyridyl)-*N*-(4-hydroxymethylcyclohexane)carboxamide (**5**) in a 57% yield. This new procedure improves the reaction yield significantly compared to the previous protocol.¹⁷

The treatment of same substrate **5** with LiAlH₄ in THF facilitated significant breakdown of the amide bond to yield compound **3** in more than 60% yield due to the possible presence of adventitious water in THF or unusual reactivity of LiAlH₄ in THF solution.¹⁷ For the preparation of the tosylated Mefway precursor **6**, the hydroxyl compound **5** was reacted with *p*-toluenesulfonyl anhydride at room temperature in dichloromethane in the presence of triethylamine to afford the precursor of [¹⁸F]Mefway in 75% yield.

In conclusion, we developed an efficient synthetic pathway for the preparation of the precursor of [¹⁸F]Mefway, which consisted of the improved the acid chloride coupling reaction to activate carboxylic acid and proper reduction condition to suppress breakdown of amide bond.

Experimental Section

***N*-2-(2-Chloroethyl)amidopyridine (1).** Chloroacetyl chloride (2.39 mL, 32.97 mmol) was slowly added to the mixture of 2-aminopyridine (1.88 g, 19.98 mmol) and TEA (4.30 mL, 30.69 mmol) in dry dichloromethane (100 mL) at 0 °C. The reaction mixture was stirred at room temperature under an Ar atmosphere for 3 h. The organic layer was extracted with CH₂Cl₂, washed with water, and dried over anhydrous MgSO₄. The residue was purified by flash column chromatography (3:1 hexane/ethyl acetate) to give product (2.68 g, 79%) as a white solid. (Caution! VERY IRRITATIVE SOLID) ¹H NMR (CDCl₃, 300 MHz) δ 4.20 (s, 2H), 7.09-7.14 (m, 1H), 7.77 (m, 1H), 8.22-8.28 (m, 2H), 9.45 (s, 1H); ¹³C NMR (CDCl₃, 75 MHz)

δ 42.8, 114.3, 120.5, 139.1, 147.1, 150.4, 164.7; HRMS (FAB⁺, *m*-nitrobenzylalcohol): Calcd for C₇H₈ON₂Cl: 171.0325, Found: 171.0324.

***N*-2-[2-{4-(2-Methoxyphenyl)-1-piperazinyl}ethyl]amido-pyridine (2).** The mixture of 1-(2-methoxy)piperazine (0.20 mL, 1.17 mmol) and K₂CO₃ (0.40 g, 2.93 mmol) in DMF (8 mL) was stirred at 80 °C for 1 h. After cooling down to room temperature, a solution of compound (1) (0.20 g, 1.17 mmol) in DMF (2 mL) and sodium iodide (0.025 g, 0.17 mmol) were added to the mixture. The reaction mixture was stirred at 80 °C for 3 h, cooled to the room temperature. The organic layer was extracted with ethyl acetate, washed with water, and dried over anhydrous MgSO₄. The residue was purified by flash column chromatography (50:1 CH₂Cl₂/MeOH to 20:1 CH₂Cl₂/MeOH) to give product (0.32 g, 84%) as pale yellow oil. ¹H NMR (CDCl₃, 300 MHz) δ 2.82-2.85 (m, 4H), 3.18 (s, 4H), 3.24 (s, 2H), 3.87 (s, 3H), 6.86-7.06 (m, 5H), 7.69-7.75 (m, 1H), 8.25-8.33 (m, 2H), 9.64 (s, 1H); ¹³C NMR (CDCl₃, 75 MHz) δ 50.6, 53.8, 55.4, 62.3, 111.2, 113.9, 118.4, 119.9, 121.0, 123.2, 138.3, 140.9, 148.0, 151.0, 152.3, 169.2; HRMS (FAB⁺, *m*-nitrobenzylalcohol): Calcd for C₁₈H₂₃O₂N₄: 327.1821, Found: 327.1818.

***N*-2-[2-{4-(2-Methoxyphenyl)-1-piperazinyl}ethyl]-*N*-(2-pyridinyl)amine (3).** 1 M LiAlH₄/THF (6.70 mL, 6.70 mmol) was slowly added to a solution of compound (2) (0.73 g, 2.24 mmol) in dry THF (10 mL) at 0 °C. The mixture was stirred at room temperature under an Ar atmosphere for 3 h. After quenching with saturated aqueous NH₄Cl at 0 °C for 30 min, the mixture was filtrated with ethyl acetate. The organic layer was extracted with ethyl acetate, washed with water, and dried over anhydrous MgSO₄. The residue was purified by flash column chromatography (50:1 CH₂Cl₂/MeOH to 20:1 CH₂Cl₂/MeOH) to give product (0.53 g, 76%) as pale yellow oil. ¹H NMR (CDCl₃, 300 MHz) δ 2.68-2.72 (m, 6H), 3.10 (s, 4H), 3.36-3.41 (m, 2H), 3.87 (s, 3H), 5.14 (s, 1H), 6.40-6.43 (d, 1H, *J* = 8.4 Hz), 6.54-6.59 (m, 1H), 6.85-7.04 (m, 4H), 7.39-7.45 (m, 1H), 8.08-8.11 (m, 1H); ¹³C NMR (CDCl₃, 75 MHz) δ 38.5, 50.7, 53.1, 55.3, 56.8, 107.0, 111.1, 112.7, 118.2, 121.0, 122.9, 137.3, 141.3, 148.2, 152.3, 158.8; HRMS (FAB⁺, *m*-nitrobenzylalcohol): Calcd for C₁₈H₂₅ON₄: 313.2028, Found: 313.2030.

***trans*-*N*-2-[2-[4-(2-Methoxyphenyl)piperazinyl]ethyl]-*N*-(2-pyridyl)-*N*-(4-carboxymethylcyclohexane)carboxamide (4).** Oxalylchloride (0.19 mL, 2.13 mmol) was added to a solution of *trans*-4-carbomethoxycyclohexane-1-carboxylic acid (0.20 g, 1.05 mmol) in dry CH₂Cl₂ (10 mL), and the mixture was refluxed for 2 h. After removing solvent and unreacted oxalylchloride in vacuo, the product was re-dissolved in dry CH₂Cl₂. Compound (3) (0.22 g, 1.05 mmol) and TEA (0.16 mL, 1.14 mmol) were added to the previous product at 0 °C. The reaction mixture was stirred at room temperature under an Ar atmosphere for 2 h. After the mixture was washed with 10% aqueous NaHCO₃ (100 mL), the organic layer was extracted with CH₂Cl₂, and dried over anhydrous MgSO₄. The residue was purified by flash column chromatography (ethyl acetate, 0.1% v/v TEA) to give product (0.28 g, 83%) as pale yellow oil. ¹H NMR (CDCl₃, 300 MHz) δ 1.17-1.25 (m, 2H), 1.62-1.66 (m, 2H), 1.84-1.96 (m, 4H), 2.25-2.29 (m, 2H), 2.58-2.63 (m, 6H), 2.98 (s, 4H), 3.62 (s, 3H), 3.84 (s, 3H), 3.95-4.00 (m, 2H), 6.83-7.00 (m, 4H), 7.24-7.31 (m, 2H), 7.74-7.80 (m, 1H), 8.51-

8.53 (m, 1H); ¹³C NMR (CDCl₃, 75 MHz) δ 27.8, 28.3, 41.3, 42.1, 44.8, 50.1, 51.4, 53.0, 55.2, 55.6, 111.0, 117.9, 120.7, 121.9, 122.4, 122.8, 138.2, 140.9, 149.2, 152.0, 175.5, 176.0; HRMS (FAB⁺, *m*-nitrobenzylalcohol): Calcd for C₂₇H₃₇O₄N₄: 481.2815, Found: 481.2814.

***trans*-*N*-2-[2-[4-(2-Methoxyphenyl)piperazinyl]ethyl]-*N*-(2-pyridyl)-*N*-(4-hydroxymethylcyclohexane)carboxamide (5).** 1 M LiAlH₄/diethyl ether (0.16 mL, 0.16 mmol) was slowly added to the solution of compound (4) (0.076 g, 0.16 mmol) in diethyl ether (5 mL) at 0 °C. The reaction mixture was stirred for 30 min at 0 °C under an Ar atmosphere, and quenched with saturated aqueous NH₄Cl. The organic layer was extracted with ethyl ether, and then evaporated solvent under reduced pressure. The residue was purified by gravity column chromatography using neutral silica gel (30:1 CH₂Cl₂/MeOH) to give product (0.041 g, 57%) as colorless oil. ¹H NMR (CDCl₃, 300 MHz) δ 1.58-1.85 (m, 10H), 2.59-2.63 (m, 6H), 2.98 (s, 4H), 3.37-3.39 (d, 2H, *J* = 6.0 Hz), 3.84 (s, 3H), 3.96-4.01 (t, 2H, *J* = 6.9 Hz), 6.83-6.98 (m, 4H), 7.22-7.28 (m, 2H), 7.74-7.76 (m, 1H), 8.51-8.53 (m, 1H); ¹³C NMR (CDCl₃, 75 MHz) δ 28.4, 28.9, 39.6, 42.3, 45.2, 50.6, 53.3, 55.3, 56.1, 68.3, 111.1, 118.1, 120.9, 122.3, 122.8, 138.1, 141.3, 149.3, 152.2, 155.8, 176.0; HRMS (FAB⁺, *m*-nitrobenzylalcohol): Calcd for C₂₆H₃₇O₃N₄: 453.2866, Found: 453.2870.

***trans*-*N*-2-[2-[4-(2-Methoxyphenyl)piperazinyl]ethyl]-*N*-(2-pyridyl)-*N*-(4-tosyloxymethylcyclohexane)carboxamide (6).** *p*-Toluenesulfonyl anhydride (0.018 g, 0.056 mmol) and TEA (0.0072 mL, 0.050 mmol) were added to the solution of compound (5) (0.021 g, 0.046 mmol) in CH₂Cl₂ (5 mL). The reaction mixture was stirred at room temperature under an Ar atmosphere for 48 h. After the mixture was washed with 10% aqueous NaHCO₃ (100 mL), the organic layer was extracted with CH₂Cl₂, and dried over anhydrous MgSO₄. The residue was purified by gravity column chromatography using neutral silica gel (20:1 CH₂Cl₂/MeOH) to give product (0.022 g, 75%) as colorless oil. ¹H NMR (CDCl₃, 300 MHz) δ 1.25-1.78 (m, 10H), 2.44 (s, 3H), 2.57-2.61 (m, 6H), 2.97 (s, 4H), 3.74-3.76 (d, 2H), 3.84 (s, 3H), 3.96-3.98 (d, 2H), 6.83-6.96 (m, 4H), 7.25-7.33 (m, 4H), 7.72-7.77 (m, 3H), 8.51-8.53 (m, 1H); ¹³C NMR (CDCl₃, 75 MHz) δ 21.6, 28.0, 28.5, 36.4, 41.8, 45.3, 50.6, 53.4, 55.3, 56.1, 74.9, 111.1, 118.1, 120.9, 122.2, 122.4, 122.8, 127.8, 129.8, 132.9, 138.2, 141.3, 144.7, 149.3, 152.2, 155.8, 175.6; HRMS (FAB⁺, *m*-nitrobenzylalcohol): Calcd for C₃₃H₄₃O₅N₄S: 607.2954, Found: 607.2954.

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